Journal of Research on the Lepidoptera 16(4):213-221, 1977

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2559 Puesta del Sol Road, Santa Barbara, California 93105

STUDIES ON THE EXCRETORY SYSTEM OF THE FULLY GROWN LARVA OF *CALOGRAMMA FESTIVA* DONOV. (NOCTUIDE)

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ABSTRACT

Fully grown larvae of *Calogramma festiva* Donov., possesses six malpighian tubules, three on either side of the gut arising from a common duct. The proximal portion remains suspended in the haemolymph as a free tubule while the distal convoluted portion of the nephric tubules in association with the rectum form the nephro-rectal complex. On the basis of histological details three regions are distinguishable in the free tubule whereas the nephric tubule is divisible into proximal and distal regions. The nephro-rectal complex is formed of nephric tubules and components from the gut wall. The pronephric epithelium and the longitudinal muscles form a cover for the nephric tubules.

INTRODUCTION

Very little is known about the excretory system of lepidopterous larvae. Ishimori (1924) has described the distribution of malpighian tubules of certain larval form of Lepidoptera. Except for the study of Mathur (1966) in noctuid caterpillars, *Prodenia litura* and *Trichoplusia ni,* no attempt has been made to investigate the structural details of malpighian tubules of lepidopterous larvae. Studies on the development of tubules in *Pieris brassicae* (Henson, 1932), *Venessa urticae* (Henson, 1937) and *Philosamia ricini* (Srivastava and Khare, 1966) give only inadequate informations on the histology of the malpighian tubules. The present work therefore, has been undertaken to describe the anatomical details of the excretory system of fully grown larva of *Calogramma festiva* D.

MATERIALS AND METHODS

Adults were collected from the field and reared in the laboratory. The fully grown larvae were anaesthesised with ether vapour and dissected in Ringer's solution under binocular microscope. For histological purposes, malpighian tubules were fixed in Bouin's fluid. Sections were obtained at 5-6 μ and staining was done with haematoxylin and eosin.

RESULTS

The excretory system of the larva of *Calogramma festiva* D., is in the form of six malpighian tubules, three on either sides of the gut. The three tubules on each side actually arise from a common duct (Fig. 1). Major portion of each tubule remains suspended in the haemolymph as free tubule while the distal portion called nephric tubule in association with the rectum forms a nephro-rectal complex.

A. *Free Tubule*

Immediately after its origin from the common duct each tubule runs towards anterior side of the gut as ascending limb (AMal) and on reaching the stomodaeum, it turns backwards and runs posteriorly as descending limb (DMal). The ascending limb of the malpighian tubule is transluscent whereas, the two thirds of the length of descending limb is dull white in colour. The remaining portion of the descending limb is characterised by the presence of small diverticulae on the wall of the tubule.

On the basis of histological details, three regions are distinguished in the free portion of malpighian tubule. In the first region (Fig. 2) peritoneal layer (Pl) is not distinct and the basement membrane (BMb) holds a ring of epithelial cells. The infoldings of the cell membrane (FcMb) in the basal zone of the cell are loosely packed and are fewer in number. In the central zone, cytoplasm is coarsely fine and contains elongated nucleus (N) measuring 5-7 μ x 2 μ, but in certain cells more elongated nuclei of 26-29 μ in size are observed. Border zone is formed of short filaments (sb) which are free towards the lumen. The second region (Fig. 3) is distinct from the first region in that the infoldings of the cell membrane (FcMb) are thickly packed in the basal zone of the cell and the reticulate cytoplasm in the central zone lodges variously shaped nucleus (N). The nucleus may be dumb-bell shaped, branched or semilunar. The cytoplasm presents scattered granules around nucleus. Border zone is formed of elogated free filaments (sb). An outer peri-

PLATE I

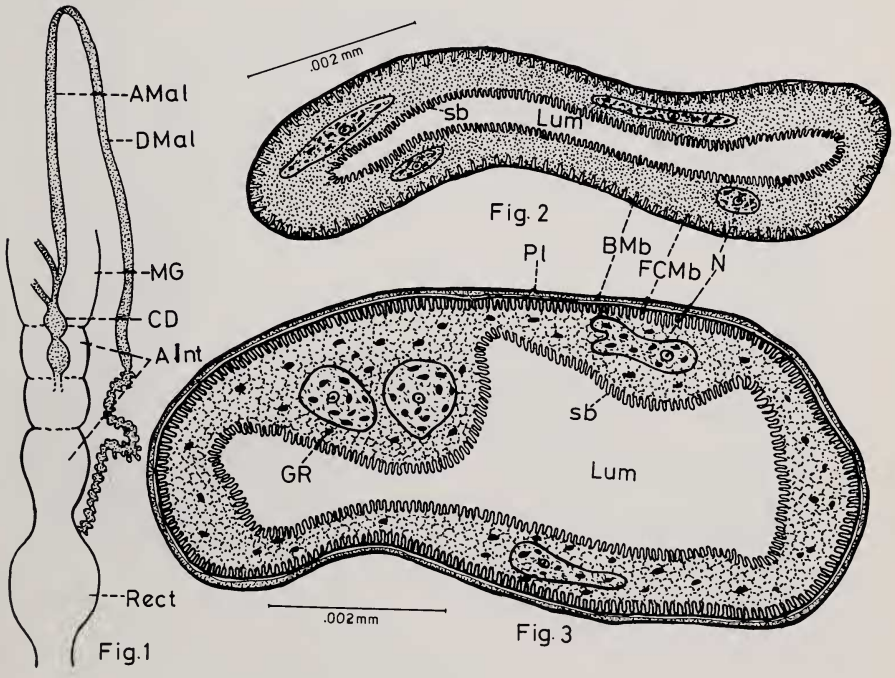

Fig. 1.—Diagrammatic representation of hind gut of *C. festiva* to show the arrangement of malpighian tubules.
Fig. 2.—Section passing through the first region of malpighian tubule of *C. festiva.*

ABBREVIATIONS

AInt, anterior intestine; AMal, ascending limb of malpighian tubule; ANt, anterior portion of nephric tubule; BMb, basement membrane; CD, common duct; DMal, descending limb of malpighian tubule; FcMb, infoldings of the cell membrane; GR, granules; In, intima; iPE, inner pronephric epithelium; Lum, lumen; mcl, muscle layer; MG, midgut; N, nucleus; oPE, outer pronephric epithelium; Pl, peritoneal layer; PNt, posterior portion of nephric tubule; RE, rectal epithelium; Rect, rectum; sb, striated border; Tra, tracheole.

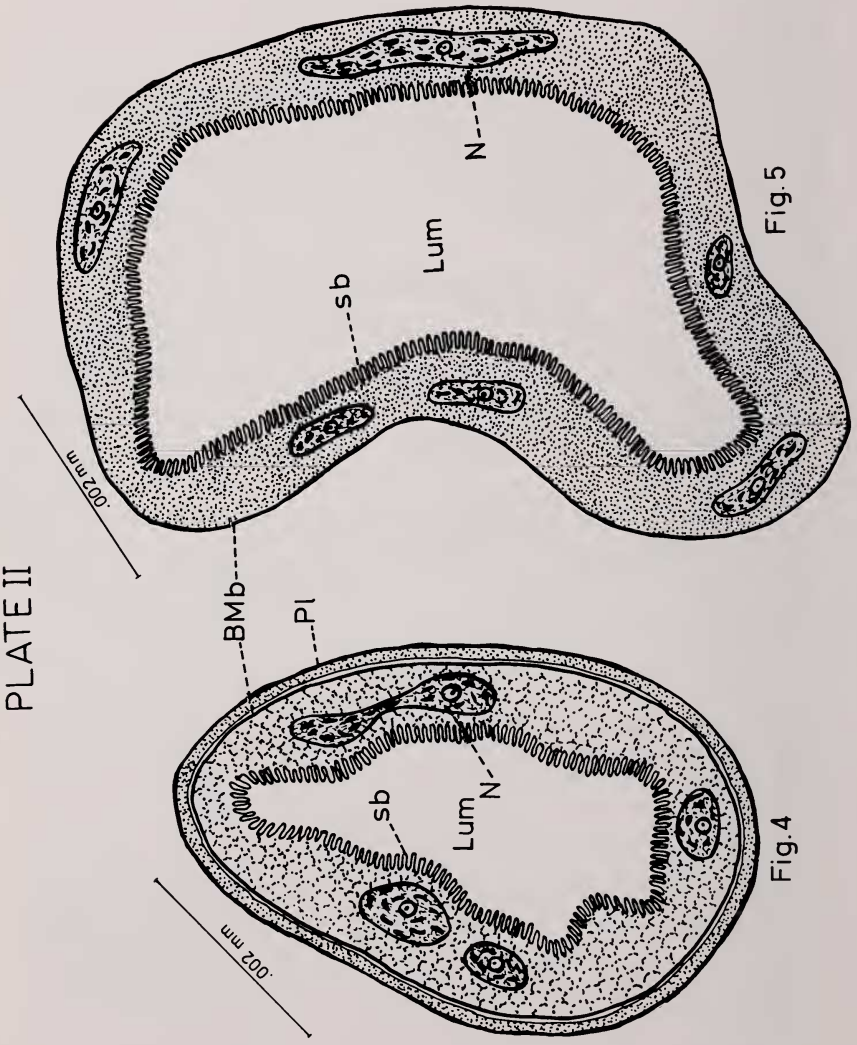

Fig. 3.—Section passing through the second region of malpighian tubule of *C. festiva.*
Fig. 4.—Section passing through the third region of malpighian tubule of *C. festiva.*

toneal layer (Pl) is present. In the third region (Fig. 4) the basal zone is highly reduced. The reticulate non-granular cytoplasm contains variously shaped nucleus (N) in the central zone. The border zone is formed of many long filaments (sb) which are free towards the lumen (Lum).

The common duct (Fig. 5) differs in histological details from the tubule. The peritoneal layer is indistinct and a tough basement membrane holds a ring of epithelial cells whose boundaries are not clear. The basal zone is highly reduced. The central zone contains dense cytoplasm. Some of the nuclei (N) present in this region are much elongated. The border zone shows elongated free filaments (sb).

B. *Nephro-rectal complex*

The nephric tubule in association with the rectum forms the nephro-rectal complex (Fig. 6) which includes the nephric tubule (Nt), pronephric epithelium (PE) and the muscle layer (mcl). The pronephric epithelium takes its origin from the gut epithelium. Immediately after its emergence from the latter, it divides into two strata, the inner pronephric epithelium (ipE) and outer pronephric epithelium (opE) both of which are formed of single layer of cells. The outer pronephric epithelium is very thin and weakly developed. The inner pronephric epithelium is well developed and formed of elongated cells. Both the layers form a loose cover over the rectal wall. Due to the provision of two epithelial layers the pronephric chamber is divided into an inner and outer chamber. Each nephric tubule after its entry into the outer chamber runs upto the posterior limit of the rectum. Thereafter it pierces the inner pronephric epithelium and enters into the inner chamber. The nephric tubule within the inner chamber takes an anteriorly directed course upto the middle of the rectum where each tubule ends blindly.

The proximal portion of the nephric tubule (ANt) lying within the outer chamber is very narrow having very thin wall. Peritoneal layer and the basal zone are indistinct. The dense cytoplasm of the central zone lodges the nucleus. The border zone is highly reduced. In the distal portion of the nephric tubule (PNt) the basal zone is formed of loosely packed infoldings of the cell membrane (FcMb). Oval nucleus is found in the reticulate cytoplasm of the central zone. The border zone is formed of long filaments (sb) which are free at their distal ends. A longitudinal muscle layer taking its origin from the gut wall surrounds the outer pronephric epithelium.

PLATE III

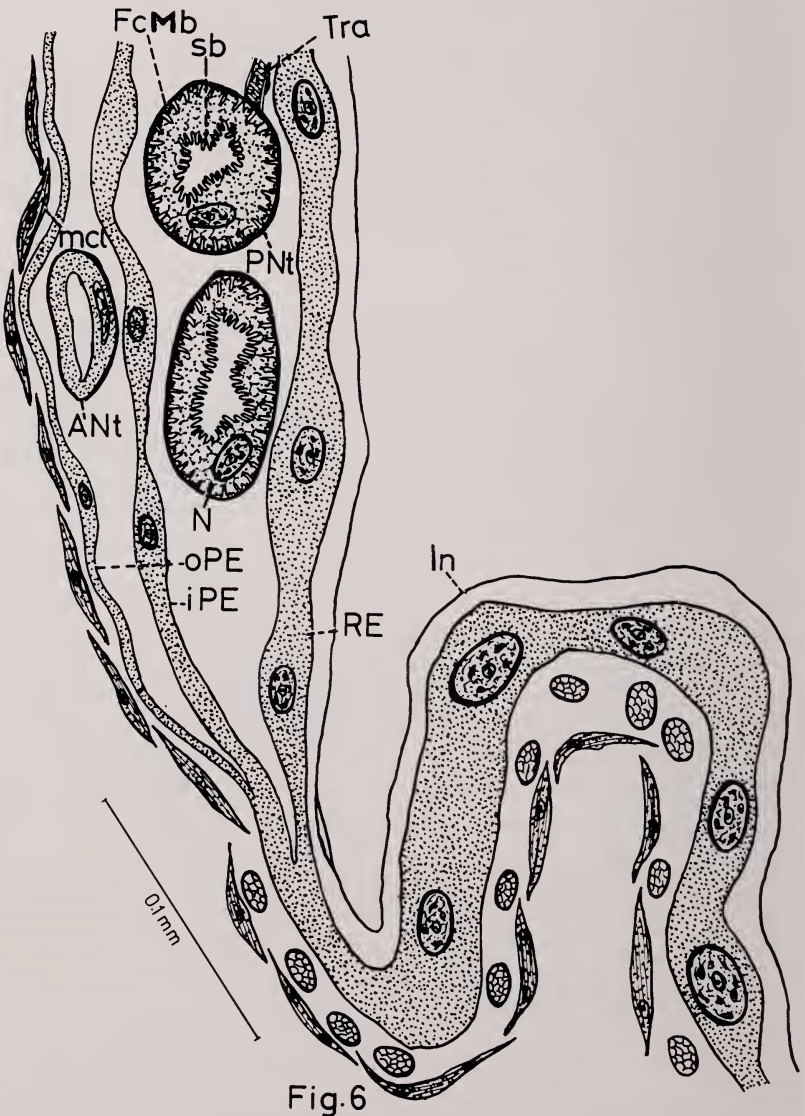

Fig. 5.—Section passing through the common duct of malpighian tubule of *C. festiva.*

Fig. 6.—Longitudinal section passing through the nephro-rectal complex of *C. festiva.*

DISCUSSION

In Lepidoptera there are six tubules in both larvae and adults (Henson, 1931; Mathur, 1966) with perhaps only exception of Tiniedae in which the six typical tubules of the larvae are reduced to only one pair in the adults (Snodgrass, 1935).

The malpighian tubules of insects are invested by a peritoneal coat having abundant supply of tracheoles (Wigglesworth, 1965). The tubules of *P. litura* and *T. ni* (Mathur, 1966) are surrounded by a peritoneal layer of very thin squamous cells with widely separated nuclei. Since there are significant variations in its structure at various levels of the tubules, it seems that these cells perhaps play a role in the tubule physiology. The nuclei of malpighian tubule differ in size, location and shape not only in various insects but also in different regions of the tubule of the same insects (Mohamed, 1974). Bertheau (1963) distinguishes five regions in the anterior part and seven regions in the posterior part of the tubule of *Carausius morosus* based on the relation between nucleus and cytoplasm. But the well marked differences between the nuclei of upper and lower segments of *Rhodnius* observed so clearly under light microscope (Wigglesworth, 1931) have not been reported in electron microscopic study (Wigglesworth and Salpeter, 1962). The size of the nucleus varies greatly in different regions of the tubule of certain aquatic bugs and many terrestrial plant feeding bugs (Bahadur, 1961; 1964). In *Forficula,* the diameter of the nuclei of the tubule at certain ecdysis become almost doubled possibly because of incomplete mitosis (Henson, 1946). The increase in the size of nucleus in *Pieris brassicae* (Henson, 1932) is accompanied by lobulation and ramification as well. So it seems evident that variously shaped elongated nuclei of *C. festiva* are either due to difference in increase of size or incomplete mitosis.

Ishimori (1924) has described the arrangement of nephric tubules in many lepidopterous larvae. The arrangement of the tubules in larval *C. festiva* is similar to that described by Henson (1931) in *V. urticae* except that instead of loops a number of convolutions are formed. In *C. festiva* the distal portion of the nephric tubule which is lodged in the inner chamber of the nephro-rectal complex extends only upto the middle of the latter as it is in the case of *Bombyx, Gastropacha* and *Spilosoma* (SAINI, 1964).

Ishimori (1924) described the 'cryptonephric envelope' of lepidopterous larvae as composed of three layers, the outer muscle layer, middle epithelial layer and inner membranous layer. The present observations agree with that of Ishimori (1924) in the number of layers but differ in the nature of pronephric epithelial layers. In the larval *Calogramma* the outer pronephric epithelial layer is very thin and weakly developed whereas, the inner pronephric epithelial layer is thick and is formed of elongated cells. It also extends firm support to the view of Srivastava and Khare (1966) that the two layers of the cells of the cryptonephric envelope originate from the epithelial layer of the gut wall.

ACKNOWLEDGEMENTS

The present work has been carried out at the Aligarh Muslim University, India and the authors are grateful to the Head of Zoology Department for providing necessary laboratory facilities. One of us (U.V.K.M.) is thankful to Prof. K. J. Joseph of the University of Calicut for enabling him to take up this work at Aligarh.

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